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BV2 Microglial Culture and Microglial Exosome Isolation

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An abbreviated version of this protocol was published in Molecular Therapy. Nucleic Acids in May 2019

Exosomes from Microglia Attenuate Photoreceptor Injury and Neovascularization in an Animal Model of Retinopathy of Prematurity

DOI: 10.1016/j.omtn.2019.04.029

Detailed protocol

BV2 microglial cells were donated by the Institute of Neurosciences at the Fourth Military Medical University. Cells were routinely cultured in DMEM (HyClone, USA) containing 10% fetal bovine serum (FBS; HyClone) at 37C. For the isolation of exosomes, microglia were cultured in FBS-free DMEM culture for 48 h, and the supernatant of the cell culture medium was collected and centrifuged at 300 g for 10 min to remove free cells. Then the supernatant was transferred into a sterile centrifuge tube. The tubes were centrifuged at 2,000 g for approximately 10 min and then at 10,000 g for 30 min to remove cell debris and cell particles. Then a 0.22-um filter (Millipore, Sigma) was used to filter the supernatant to remove particles. Ultracentrifugation was used to isolate exosomes at 100,000 g for 70 min. We collected the pelleted exosomes, washed them with PBS, centrifuged them again at 100,000 g for 70 min, and resuspended the pellet in 100 uL of PBS. All procedures were conducted at 4C. Exosomes were stored at 80C for less than 1 week or used immediately for downstream experiments.

How to cite:(Readers should cite both the Bio-protocol preprint and the original research article where this protocol was used)

1. Wang, Y. (2021). BV2 Microglial Culture and Microglial Exosome Isolation. Bio-protocol Preprint. bio-protocol.org/prep1192.
2. Xu, W., Wu, Y., Hu, Z., Sun, L., Dou, G., Zhang, Z., Wang, H., Guo, C. and Wang, Y.(2019). Exosomes from Microglia Attenuate Photoreceptor Injury and Neovascularization in an Animal Model of Retinopathy of Prematurity. Molecular Therapy. Nucleic Acids 16. DOI: 10.1016/j.omtn.2019.04.029